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Molecular Characterization of Peptoid Nanosheet Assembly at the Liquid/Liquid Interface

Facilities

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Foundry Facilities

Facility	Description
Biological: (lead)	We require the resources of the Biological Nanostructures Facility at the Molecular Foundry, as well as the expertise of the researchers, to provide peptoid samples for our spectroscopic measurements via custom peptoid design, synthesis, purification, and characterization. Specifically, we require Foundry capabilities for routine peptoid synthesis, purification and characterization by HPLC. We also require complementary monolayer characterization using both Langmuir trough experiments to obtain surface pressure isotherms as well as fluorescence microscopy, as well as characterization of the nanosheets with optical imaging.
Imaging/Manipulation: (support)	Additional capabilities that the Molecular Foundry provide that are important for the success of this project is the access to instrumentation in the Imaging and Manipulation of Nanostructures Facility. Specifically, we require peptoid nanosheets produced from the oil-water interface to be characterized by routine AFM and SEM measurements.
Theory: (support)	Because VSF spectra provide a wealth of information related to the number, orientation, solvation, and bonding of interfacial molecules, it can be difficult to de-convolute these factors. While complicated fitting routines are employed to interpret the spectra of peptoid polymers adsorbed to the oil-water interface, molecular modeling of peptoid monolayers can be very helpful in providing additional insight into the molecular level details of this key intermediate. We therefore require expertise from the researchers in the Theory Facility, whose efforts have already proven successful in simulating peptoid polymers, to model peptoid monolayers at the oil-water interface.

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Proposal Description

Significance and Impact

Peptoid nanosheets are a novel class of two dimension nanomaterials that form from the assembly of peptoid polymers.¹ The Zuckermann group at the Molecular Foundry has discovered that the air-water interface catalyzes the formation of nanosheets.² Specifically, the mechanism for nanosheet synthesis first involves the assembly of a peptoid monolayer at the interface, with subsequent compression leading to monolayer collapse and formation of stable bilayer structures. Recently, nanosheet formation has also been shown to occur via the oil-water interface, a unique platform for synthesis that has the potential to increase nanosheet functionality and stability.³

In order to increase the complexity and functionality of these novel nanomaterials, as well as to better explain the mechanism of formation, it is essential to understand the molecular level details of the key peptoid monolayer intermediate that either contribute to or inhibit nanosheet formation. A depiction of the monolayer is shown in shown in Figure 1. In particular, it is important to understand the structural requirements of peptoids at the interface (such as degree of backbone and side chain ordering, intermolecular interactions, and packing density) required to make nanosheets.

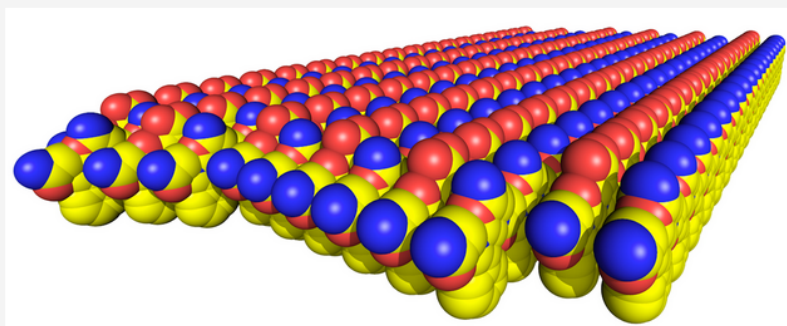


Figure 1. Cartoon of a peptoid monolayer.

One way of examining this adsorption and structural ordering is to use the technique vibrational sum frequency (VSF) spectroscopy that is a well-established surface selective technique that produces vibrational spectra of oriented interfacial molecules.⁴ This technique provides fundamental information related to bond strengths, interactions between chemical species, and orientations of molecules specifically at the interface. Based on these characteristics, VSF spectroscopy is an ideal technique to probe the atomistic details of peptoid monolayers at the oil-water interface. We therefore propose to study monolayers of a variety of sequence specific peptoids at the oil-water interface, in which the ability or inability of the peptoid to form nanosheets is known. By understanding the role of each molecular moiety of the peptoid in interfacial assembly and subsequent nanosheet formation, peptoid polymers can be better designed to more effectively assemble into nanosheets for a variety of interesting applications that include molecular sensors⁵, artificial membranes, and catalysts.

References

- Nam, K. T.; Shelby, S. A.; Choi, P. H.; Marciel, A. B.; Chen, R.; Tan, L.; Chu, T. K.; Mesch, R. A.; Lee, B.-C.; Connolly, M. D.; Kisielowski, C.; Zuckermann, R. N. Free-Floating Ultrathin Two-Dimensional Crystals from Sequence-Specific Peptoid Polymers. *Nat. Mater.* **2010**, *9*, 454-460.
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- Olivier, G. K.; Cho, A.; Sanii, B.; Connolly, M. D.; Tran, H.; Zuckermann, R. N. Antibody-Mimetic Peptoid Nanosheets for Molecular Recognition. *ACS Nano* **2013**, *7*, 9276-9286.

Project Plan

The proposed project to understand the molecular level details of peptoid monolayers at the oil-water interface combines the synthetic capabilities of Ron Zuckerman's group in the Molecular Foundry and the VSFS capabilities of [redacted] group at the [redacted]. Over the past year initial collaborative studies have resulted in some important results that have been submitted to *PNAS*.¹ The studies demonstrate the highly ordered nature of the monolayers of two different peptoids at the oil-water interface. Their chemical structures are shown in Figure 2.

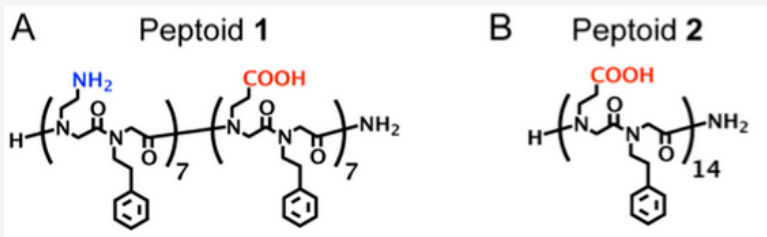


Figure 2. Chemical structures of peptoids used in previous VSF spectroscopic study.

At pH 8, Peptoid 1 was able to form nanosheets, while Peptoid 2 was not. Through our VSF spectroscopic and interfacial tension studies, we discovered that electrostatic interactions between positively charged amine and negatively charged carboxylate groups are key in the ability for Peptoid 1 to form a highly packed monolayer at the oil-water interface, which contributes to the ability of the monolayer to collapse and form nanosheets. Charge-charge repulsions in monolayers of Peptoid 2 hinder ordered interfacial assembly, which contributes to the inability of this peptoid to form nanosheets.

Since favorable electrostatic interactions appear to be so important, we propose to study the role of metal cations (Ca^{2+} , Mg^{2+} , Mn^{2+} , and Cu^{2+}) in the ability of carboxylic acid containing peptoids, such as Peptoid 2, to form nanosheets via the oil-water interface. VSF spectroscopic measurements will be used to determine how the molecular level details of the monolayer at the oil-water interface relate to the ability of the peptoid to form nanosheets. We will specifically look for electrostatic interactions between the carboxylate group and the metal ions by probing the carboxylate stretching mode near 1400 cm^{-1} . Observing the specific peak frequency will allow us to determine whether the interactions between the metal ions and the carboxylate groups are purely electrostatic in nature, or if the carboxylate groups are able to chelate the metal ions.

We will also explore the role of the bulkiness hydrophobic moieties by studying peptoids that contain different ethyl phenyl groups, as shown in Figure 3. By probing the CH moieties of the adsorbed peptoid backbone, side chains, and aromatic groups, we will be able to obtain information related to the packing ability of these peptoids at the oil-water interface and how this degree of packing relates to the ability to form nanosheets.

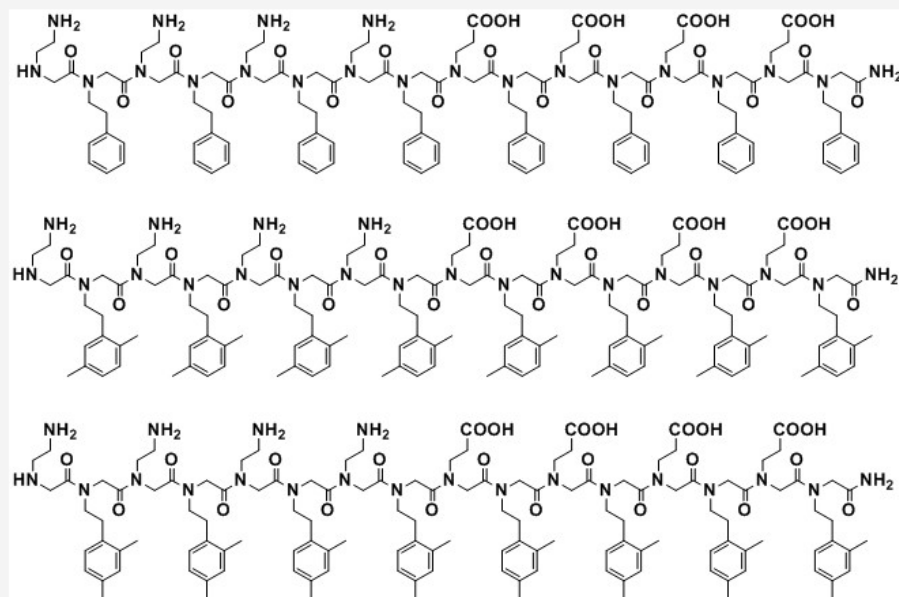


Figure 3. Chemical structures of peptoids in which the bulkiness of the phenyl group is varied.

For these projects, peptoid synthesis and characterization, nanosheet formation via the oil-water interface, and nanosheet characterization will be performed at the Molecular Foundry. VSF spectroscopic measurements, along with complementary interfacial tension studies, will be performed at the [redacted].

References

1. [redacted]
Pep

Molecular Foundry Utilization Timeline

We expect that the time to collect the VSF spectroscopic and interfacial tension data required for these project to take approximately 1 year. These measurements will be performed by Dr. [REDACTED] at the [REDACTED], and so the time needed from the resources at the Molecular Foundry will mainly involve routine peptoid synthesis and purification, as well as nanosheet formation and characterization. We anticipate needing less than 5 new peptoids on a 10 mg scale to be sufficient to drive these studies. The spectroscopic and interfacial tension measurements require very little sample (10 mL of 100?M peptoid). Dr. [REDACTED] may also periodically need to visit the Molecular Foundry in order to perform additional characterizations of the peptoid monolayers at the oil-water interface using Langmuir trough experiments and fluorescence microscopy. We expect him to use these facilities once every three months.

Relevant Experience

Directing the effort is Prof. [REDACTED] who is a well recognized for her many research contributions to the molecular understanding of interfacial assembly and adsorption at complex liquid interfaces using VSFS, theoretical simulations and complementary thermodynamic methods. This project builds on this expertise while expanding her effort to more complex biomolecular systems. She is a member of the NAS, American Academy of Arts and Sciences and a Fellow of the APS and ACS.

Dr. [REDACTED], a post-doc in the [REDACTED] lab at the [REDACTED] since 2013 will conduct the experiments. Dr. [REDACTED] is well skilled in the VSF technique through his related studies of both surfactants and nanoparticles at the liquid-liquid interface. He has considerable additional experience in the biophysical chemistry area through his Ph.D. work with Prof. [REDACTED] at the [REDACTED]. Dr. [REDACTED] plans to report the findings from the peptoid collaboration between the [REDACTED] and Zuckermann labs at the next American Chemical Society National meeting in San Francisco.

Need for the Molecular Foundry

The Molecular Foundry provides unique capabilities that are not available at the [REDACTED] or the [REDACTED] Laboratory. With the focus on surface spectroscopic measurements of simpler surfactant and polymer systems at liquid surfaces, the [REDACTED] laboratory is well equipped with various VSF laser stations for doing the experiments and equipment for measuring interfacial tension. The laboratory however is not equipped for performing the synthesis of peptoid polymers nor does it have any expertise in peptoid synthesis and characterization. Of particular importance in these proposed studies is the need to have experts to synthesize peptoids of extremely high purity. Fortunately, several high purity molecules needed for the spectroscopic and interfacial tension measurements are already known and available from the Molecular Foundry.¹⁻³ For new peptoid polymers, we require the resources at the Molecular Foundry, as well as the expertise of the researchers, for providing us with peptoid samples to be used in the spectroscopic and interfacial tension measurements at the [REDACTED].

Because our goal is to relate molecular level characteristics of peptoid monolayers to the ability of peptoid to form nanosheets, it is essential to be able to determine whether or not nanosheets form from specific peptoids. At the [REDACTED], we neither have the capability or expertise to prepare peptoid nanosheets in a controlled manner from the oil-water interface, nor access to methods used for nanosheet characterization. We therefore require resources at the Molecular Foundry to prepare and characterize nanosheets made via the oil-water interface from peptoids that we will study with VSF spectroscopy.

References

1. Nam, K. T.; Shelby, S. A.; Choi, P. H.; Marciel, A. B.; Chen, R.; Tan, L.; Chu, T. K.; Mesch, R. A.; Lee, B.-C.; Connolly, M. D.; Kisielowski, C.; Zuckermann, R. N. Free-Floating Ultrathin Two-Dimensional Crystals from Sequence-Specific Peptoid Polymers. *Nat. Mater.* **2010**, *9*, 454-460.
2. Sanii, B.; Kudirka, R.; Cho, A.; Venkateswaran, N.; Olivier, G. K.; Olson, A. M.; Tran, H.; Harada, R. M.; Tan, L.; Zuckermann, R. N. Shaken, Not Stirred: Collapsing a Peptoid Monolayer to Produce Free-Floating, Stable Nanosheets. *J. Am. Chem. Soc.* **2011**, *133*, 20808-20815.
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